APPENDIX N: REUSE OF A POLYACRYLAMIDE ELECTROPHORESIS GEL

1 EQUIPMENT

1.1 SA-43 vertical electrophoresis tank

1.2 Power supply

2 MATERIALS

2.1 Kimwipes

2.2 Disposable 60 cc syringe

2.3 Disposable, large bore transfer pipettes

2.4 Plastic wrap

2.5 Paper towels

3 REAGENTS

3.1 Ethanol, 95%

3.2 1X TBE buffer

4 PROCEDURE

4.1 After the gel has been scanned in the FMBIO, place it into the SA-43 vertical electrophoresis tank.

4.2 Secure the gel in place and add 1X TBE buffer to the upper and lower tank reservoirs.

4.3 To run the previously loaded samples out of the top of the gel, connect the leads from the power supply so that the red (+) electrode is attached to the top black electrode and the black (-) electrode is attached to the red lower electrode of the electrophoresis tank.

4.4 Turn the power supply on and begin to electrophoresis at 60 Watts for a minimum of 2½ hours. It is recommended that the typing gel be run in the reverse direction for approximately ½ hour longer than the typing gel was run in the forward direction. This will ensure that all of the DNA has run out of the top of the gel prior to loading new samples.

NOTE: If the DNA samples are not run out of the top of the gel on the same day as they were loaded, wrap the typing gel in plastic wrap with a moistened paper towel wrapped around the top, bottom, and sides of the gel and then store at room temperature in the dark until the reverse electrophoresis is conducted.

4.5 Once the samples have run out of the top of the gel into the top tank buffer reservoir, drain the upper tank buffer reservoir. Remove the gel from the SA-43 electrophoresis tank and discard the tank buffer in the lower buffer reservoir.

4.6 Rinse the upper tank reservoir with water and wipe with a Kimwipe to remove any remaining buffer.

NOTE: If the gel will not be reused the same day, wrap it in plastic wrap with a moistened paper towel wrapped around the top, bottom, and sides of the gel and then store at room temperature in the dark.

4.7 Place the gel into the SA 43 vertical gel electrophoresis tank, large plate 'facing out', and turn the clamps to secure the gel in place. Add 1X buffer to the upper reservoir of the gel tank. Flush the wells with the tank buffer using a syringe or a large bore transfer pipet.

NOTE: Electrophoresis gels should not be used more than 3 times.

4.8 Fill the lower reservoir of the gel tank with 1X TBE buffer. Remove any air bubbles that are trapped between the bottom of the gel and the buffer in the lower reservoir. Then proceed with steps 7.5.10 through 7.5.20 for PowerPlex® 16 BIO System typing gels, steps 7.6.10 through 7.6.19 for PowerPlex® 1.1 System typing gels, and steps 7.7.10 through 7.7.19 for PowerPlex® 2.1System typing gels.

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